65. The plant according to claim 58, wherein the expression cassette further comprises the terminator of the tobacco PR-1a gene operably linked downstream of the DNA sequence encoding the member of the sarcotoxin 1 family or the cecropin family.

66. The plant according to claim 58, wherein the constitutively expressed promoter is the cauliflower mosaic virus 35S promoter.

67. The plant according to claim 58, wherein the expression vector further comprises a T-DNA region and a drug resistance gene.--

## REMARKS

**Status**

Claims 48-67 are pending in this application after entry of this amendment, with claims 48-67 being added, claims 21-47 being canceled and no claims being amended herein. Support for the new claims may be found at least in the claims as filed on February 23, 2000 and in the specification as follows. The new claims recite "a member of the sarcotoxin 1 family or the cecropin family of antibacterial peptides from a Diptera insect". Support for these new claims may be found in the specification at least on page 2, lines 9-18 and page 10, lines 1-17. Claims 48, 52, and 58 recite the components of the recombinant vectors used to generate the claimed transgenic plants. Support for these new claims may be found in the specification at least on page 2, lines 9-18; page 4, lines 16-25, page 10, line 24 - page 11, line 3; and page 11, line 32 - page 12, line 33. Claim 53 contains the correct spelling of chitinase. Claim 58 recites "promoter induced by stress" instead of "inducible promoter". Support for this new claim may be found in the specification at least on page 9, lines 6-8. Claims 48 and 58 recite "a corresponding untransformed plant" as suggested by the Examiner in the office action dated March 9, 2001. No new matter is added by these new claims.

The Advisory Actions mailed September 14, 2001 and October 16, 2001 indicated that the amendments filed August 23, 2001, September 18, 2001 and September 21, 2001 were not entered on the grounds that they raised issues that would require further consideration and/or search and they did not put the application in better form for appeal. The request for reconsideration was considered but deemed not to overcome the rejections stated in the March 9, 2001 Office Action. In that action, prior claims 21, 22, 24-33, and 35-47 were rejected under 35 U.S.C. §112, first paragraph on the grounds that Applicants did not have possession of the claimed invention at the time of filling the application. Prior claims 21, 22, 24-33, and 35-47 were rejected under 35 U.S.C. §112, first paragraph as allegedly not enabled. Prior claims 21-47 were rejected under 35 U.S.C. §112, second paragraph on the grounds they are indefinite. Applicants respectfully traverse these rejections and address these rejections as they relate to the new claim set filed herewith.

**35 U.S.C. §112, first paragraph, possession:**

Prior claims 21, 22, 24-33, and 35-47 were rejected under 35 U.S.C. §112, first paragraph on the grounds that they contain subject matter which was not described in the specification in such a way as to reasonably convey to one of skill in the art that the inventors, at the time the application was filed, had possession of the claimed invention. In particular, the Examiner alleged that Applicants had not satisfactorily described the claimed genus of nucleic acids required to practice the claimed invention. Applicants respectfully disagree.

Recently, the Court of Appeals for the Federal Circuit ("CAFC") stated that "[i]n written description cases, 'the primary consideration is factual and depends on the nature of the invention and the amount of knowledge imparted to those of skill in the art by the disclosure.'" *Union Oil Co. v. Atlantic Richfield Co.*, 208 F.3d 989, 996 (Fed. Cir. 2000), quoting *In re Wertheim*, 541 F.2d 257, 262 (CCPA 1976). The CAFC also stated:

> **The written description requirement does not require the applicant 'to describe exactly the subject matter to be claimed**, instead the description must clearly allow persons of ordinary skill in the art to recognize that he or she invented what is claimed.'" [emphasis added] *Id.* at 997, quoting *In re Gosteli,* 872 F.2d 1008, 1012 (Fed. Cir. 1989).

The CAFC further stated:

> Rather, the Patent Act and this court's case law require only sufficient description to show one of skill in the refining art that the inventor possessed the claimed invention at the time of filing." *Id.*

In light of this case law, it is clear that the question to be resolved is whether the present patent application provides sufficient description to allow those of skill to recognize what has been invented. The invention claimed here is not the identification of genes encoding the antibacterial peptides from the Diptera insect. The invention is the use of such genes in new methods that confer new properties on plants.

Although Applicants disagree with the Examiner's position, in view of the Examiner's position, Applicants provide new claims to limit the scope of the genus to genes encoding anti-bacterial peptides that are "a member of the sarcotoxin 1 family or the cecropin family of antibacterial peptides from a Diptera insect". These amendments clarify the scope of the claimed invention. Applicants claim:

> A method of conferring resistance to pathogenic fungi on a plant using a DNA sequence encoding a member of the sarcotoxin 1 family or the cecropin family of antibacterial peptides from a Diptera insect, the method comprising the steps of: transforming a plant cell by introducing the DNA sequence encoding the member of the sarcotoxin 1 family or the cecropin family; and regenerating the transformed plant cell into a transgenic plant expressing the member of the sarcotoxin 1 family or the cecropin family, wherein the DNA encoding the member of the sarcotoxin 1 family or the cecropin family from a Diptera insect is in an expression vector, wherein said expression vector comprises:

i) an expression cassette comprising a first plant promoter induced by stress; and
ii) a second plant promoter which is constitutively expressed,
wherein the first plant promoter and the second plant promoter are positioned adjacent to each other, and wherein the transgenic plant has enhanced resistance to pathogenic fungi as compared to a corresponding untransformed plant.

Applicants teach methods of constructing a recombinant gene and an expression cassette (Examples 1 and 2), methods of transforming a plant cell (Example 3), generating plants with enhanced resistant to pathogenic fungi compared to untransformed plants (Examples 5-6, and 8-10). Furthermore, Applicants provide a working example wherein a transgenic plant comprising a member of the sarcotoxin 1 family of antibacterial peptides is made. Applicants show that the transgenic plant has enhanced resistance to pathogenic fungi compared to untransformed plants as claimed.

As discussed in the previous responses filed February 2, 2001 and August 23, 2001 and including the references cited and enclosed therein, the sarcotoxin 1 family of peptides were well known in the art prior to filing the above referenced application. The sarcotoxin 1 family of peptides include sarcotoxins 1a, 1b, 1c, 1d, and the cecropin family of peptides include cecropins A, B, C, and D. These peptides are short and nucleic acids encoding these peptides were also well known to those of skill in the art prior to filing the above referenced application. For example, Kylsten *et al. EMBO Journal* 9: 217-224 (1990) disclose the nucleotide sequence of the *Drosophila* cecropins A1, A2, and B (see Figure 2, page 218) and compare the predicted amino acid sequences to sarcotoxin 1a (see Figure 3, page 219). Matsumoto *et al., Biochem. J.* 239: 717-722 (1986) describe the cloning of and report the nucleotide sequence encoding the sarcotoxin 1a peptide from *Sarcophaga peregrina* (see Figure 1, page 719). Kanai *et al., FEBS Lett.* 258: 199-202 (1989) describe the cloning of a gene cluster encoding the sarcotoxin 1 family and report the nucleotide sequence encoding sarcotoxin 1b from *Sarcophaga peregrina* (see Figure 1, page 200 and Figure 5, page 201). Okada *et al., J. Biological Chem.* 260: 7174-7177 (1985) cited in the February 2 response, disclosed that the

sarcotoxin 1c amino acid sequence differed from the sarcotoxin 1a amino acid sequence by only two amino acids (see Figure 5, page 7176). One of skill in the art using the known genetic code would be able to generate a nucleotide sequence encoding the sarcotoxin 1c peptide based on the sarcotoxin 1a nucleotide sequence using known PCR mutagenesis techniques. Based on the extensive homology of the sarcotoxin 1 and cecropin peptides in the Diptera insects, one of skill would be able to generate a nucleotide sequence encoding any known sarcotoxin or cecropin peptide using known PCR mutagenesis techniques.

The Examiner admits on page 3 of the office action dated March 9, 2001, that references cited previously provide written description of cecropin nucleic acids from *Drosophila* and *Ceratitis* species. The Examiner also states on page 3 of the office action "[w]hereas Applicant need not disclose all of the species encompassed by the claimed genus, Applicant must describe a representative number of species."

In view of the above remarks, Applicants submit that they have provided an adequate written description of a sufficient number of species of the genus as currently claimed, as required under 35 U.S.C. §112, first paragraph, and that Applicants were in possession of the claimed invention at the time the application was filed.

**35 U.S.C. §112, first paragraph, enablement:**

Prior claims 21, 22, 24-33, and 35-47 were rejected under 35 U.S.C. §112, first paragraph on the grounds that the specification is enabling only for claims limited to methods of enhancing fungal resistance with, and transgenic plants comprising, the sarcotoxin 1a gene. In particular, the Examiner states Applicants have not provided evidence that the structural relatedness of the sarcotoxin, and cecropin peptides corresponds to functional relatedness and that a cecropin peptide can be substituted for *e.g.* a sarcotoxin peptide. The Examiner further states that undo experimentation would be required to produce the nucleic acid molecules encoding the sarcotoxin family of peptides. Applicants respectfully disagree.

It is well settled that the test of enablement is whether one reasonably skilled in the art could make or use the invention from the disclosures in the patent coupled with information known in the art without undue experimentation. See *In re Wands*, 8 USPQ2d at 1404 (Fed. Cir. 1988) and MPEP §2164.01. It is also well settled that the Examiner bears the burden of showing why an application fails to provide an enabling disclosure. *In re Marzocchi*, 169 USPQ 367 (CCPA 1971). Applicants respectfully submit that based on the evidence provided, one of skill could easily prepare constructs using nucleotide sequences encoding sarcotoxins other than sarcotoxin 1a or nucleotide sequences encoding cecropins, make transgenic plants and easily determine the extent to which they enhance the resistance to pathogenic fungi. The fact that such additional routine testing may be required to identify plants within the scope of the invention is not dispositive. As noted by the courts, the test of enablement is not whether any experimentation is necessary, but whether, if experimentation is necessary, it is undue. *In re Angstadt*, 190 USPQ 214, 219 (CCPA 1976).

The Examiner is reminded:

> Enablement is a legal determination of whether a patent enables one skilled in the art to make and use the claimed invention, *Raytheon Co. v. Roper Corp.*, 724 F.2d 951, 960(Fed. Cir. 1983), is not precluded even if some experimentation is necessary, although the amount of experimentation needed must not be unduly extensive, *Atlas Powder Co. v E.I. Du Pont De Nemours & Co.*, 750 F.2d 1569, 1576 (Fed. Cir. 1984), and is determined as of the filing date of the patent application. . . *See W.L. Gore and Associates v. Garlock, Inc.*, 721 F.2d 1540, 1556 (Fed. Cir. 1983). Furthermore, a patent need not teach and preferably omits, what is well known in the art. *Lindemann,* 730 F.2d at 1463, *Hybritech v. Monoclonal Antibodies, Inc.* 802 F.2d 1367, 1374 (Fed. Cir. 1986).

As the Examiner admitted on page three of the March 9, 2001 Office Action, the specification is enabling for methods of enhancing fungal resistance with, and transgenic plants comprising the sarcotoxin 1a gene. As discussed above, in view of the Examiner's comments, Applicants submit new claims herewith limiting the scope of the claims to the sarcotoxin 1 family and cecropin family of antibacterial peptides from the

Diptera insect. As discussed above, Applicants had submitted references in the response filed January 26,2001, indicating that genes encoding antibacterial peptide from Diptera insects including genes encoding the sarcotoxin 1 family and the cecropin family were well known in the art. In the response filed August 23, 2001, Applicants submitted additional references containing sequence information indicating that nucleic acid sequences encoding members of the sarcotoxin 1 family or cecropin family were well known to those of skill in the art prior to filing the above referenced application or could be obtained without undue experimentation using well known techniques.

Applicants also enclosed additional references in the August 23, 2001 response indicating that the structural similarity among the sarcotoxin 1 family members and the cecropin family members relate to functional similarities as well. For example, Iwai *et al., Eur. J. Biochem.* 217: 639-644 (1993) disclose the NMR structure of sarcotoxin 1a. Data presented indicate that sarcotoxin 1a consists of 2 amphiphilic α-helices (see Figures 1, 5, and 6 and page 643, right column, second, third and fifth full paragraphs), which is suggested to be important for penetrating the bacterial membrane. Iwai *et al.,* cite to a similar structural analysis of cecropin A, which indicates that cecropin A also contains two helical regions located in the same portions of the peptide as the sarcotoxin 1a helices (see page 643 right column, fifth paragraph and Figure 7). Iwai *et al.,* suggest that the structure of cecropin B is similar to cecropin A.

Other references provided indicate that the cecropins, like the sarcotoxins, also have a fungicidal activity. For example, DeLucca *et al., Antimicrobial Agents and Chemotherapy* 41: 481-483 (1997), cited in the February 2, 2001 response, reported that cecropin A had fungicidal activity against both *Aspergillus* and *Fusarium* species (see page 482, left column and Figures 1 and 2). DeLucca *et al., Medical Mycology* 36: 291-298 (1998) further report that cecropin B also had fungicidal activity against both *Aspergillus* and *Fusarium* species (see page 293, right column, page 294, Figure 1, and page 295, Discussion, paragraphs 1-3). Ekengren *et al., Insect Biochemistry and Molecular Biology* 29: 965-972 (1999) report that cecropin A from *Drosophila* or *Hyalophora* and cecropin B from *Drosophila* were fungicidal against a number of

different fungi (see page 967, paragraph 3.1 and Figure 1 and page 968, left column end of continued paragraph, right column beginning of Discussion and Table 1).

As discussed above, nucleic acids encoding the sarcotoxin 1 family and cecropin family of antibacterial peptides were also known or easily generated using known techniques by those of skill in the art the time the application was filed (see *e.g.* Matsumoto *et al., Biochem. J.* 239, 717-722 (1986), Kanai *et al., FEBS Lett.* 258: 199-202 (1989) and previously cited Figure 2 of Kylsten *et al., The EMBO J.* 9: 217-224 (1990), and Figure 1 of Rosetto *et al., Gene* :134: 241-243 (1993)). For example, one of skill in the art could easily modify the known nucleic acid sequence of sarcotoxin 1a to produce a nucleic acid sequence encoding sarcotoxin 1c or 1d based on the known genetic code and the ability to synthesize long oligonucleotides for use in PCR reactions. Because the nucleotide sequences and/or amino acid sequences of various sarcotoxin family members are known, one of skill would not need to screen through a vast array of degenerate nucleic acid molecules, as the Examiner suggests, to generate nucleotide sequences encoding a sarcotoxin 1 family member or a cecropin family member. As stated above, "[f]urthermore, a patent need not teach and preferably omits, what is well known in the art." *Id.*

The Examiner suggests that the application is not enabled because Florack *et al., Transgenic Research* 4: 132-141 (1995) report constructing a transgenic tobacco plant containing cecropin B which failed to confer resistance to pathogenic bacteria. In discussing the Florack *et al.,* article in their response filed December 28, 1999, Applicants distinguished the instant application from Florack stating in the first full paragraph on page 10 of the response "the plant expression system used by Florack *et al.,* was probably not efficient enough to express an amount of peptide necessary to confer resistance to pathogenic bacteria on the plant." Applicants describe a dual promoter system on page 9, lines 5-17 of the specification and claimed in claim 48, lines 9-11 wherein both a constitutively expressed promoter and a promoter induced by stress control the expression of the sarcotoxin 1 family member or the cecropin family member. Florack *et al.,* reported using only a constitutively expressed promoter to drive expression

of cecropin B in their transgenic tobacco plants. Moreover, Applicants provide working Examples 5 and 6 and Figures 10 and 11 showing that the expression system used in the instant application is able to produce a sufficient amount of peptide in a transformed plant to convey enhanced resistance to pathogenic fungi compared to a corresponding untransformed plant as claimed. As stated in §2164.03 of the MPEP:

> For a claimed genus, representative examples together with a statement applicable to the genus as a whole will ordinarily be sufficient if one skilled in the art (in view of level of skill, state of the art and the information in the specification) would expect the claimed genus could be used in that manner without undue experimentation. Proof of enablement will be required for other members of the claimed genus only where **adequate reasons** are advanced by the examiner to establish that a person skilled in the art could not use the genus as a whole without undue experimentation. [emphasis added]

Due to the differences in the expression systems described in the instant application and Florack *et al.*, and the presence of working examples, Applicants submit that the Examiner has not advanced adequate reasons to establish that a person skilled in the art could not use the genus as a whole without undue experimentation.

In view of the above remarks, Applicants submit that the currently pending claims are fully enabled as required by 35 U.S.C. §112, first paragraph.

**35 U.S.C. §112, second paragraph, indefiniteness:**

Prior claims 21-47 were rejected under 35, U.S.C. §112, second paragraph as allegedly indefinite. In particular, prior claims 21 and 32 were rejected on the grounds that the phrase "compared to the plant before transformation" in lines 6-7 or 7 respectively is unclear because the plant did not exist prior to transformation. Applicants have adopted the Examiner's suggestion and have drafted new claims to recite -- compared to a corresponding untransformed plant--.

Prior claim 25 was rejected on the grounds that the word "chitinasa" was misspelled. Applicants have adopted the Examiner's suggestion and corrected the spelling in new claims 53 and 62.

Prior claims 42-47 were rejected as allegedly indefinite for containing an improper Markush group and for lacking antecedent basis for the term "Diptera insect". Prior claims 42, 43, 45, and 46 were rejected because the term homolog is allegedly indefinite. Prior claims 42 and 45 were rejected because the phrase "derived from" is indefinite. Applicants have canceled claims 42-47 herein and have drafted new claims in view of the Examiner's remarks.

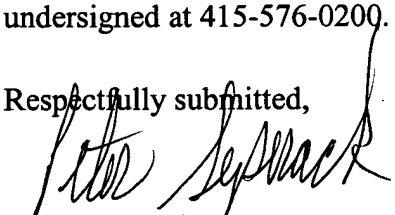
Prior claims 22-24, 26-31 and 33-41 were also rejected although the Examiner did not specifically enumerate a reason for the rejection. Applicants submit that, in view of the above comments, all new claims comply with the requirements of 35 U.S.C. §112, second paragraph.

## CONCLUSION

In view of the foregoing, Applicants believe all claims now pending in this Application are in condition for allowance. The issuance of a formal Notice of Allowance at an early date is respectfully requested.

If the Examiner believes a telephone conference would expedite prosecution of this application, please telephone the undersigned at 415-576-0200.

Respectfully submitted,



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